



Serial No.: 10/768,996  
Shrivastava et al.

Art Unit: 1642

## IN THE CLAIMS

Please replace prior versions and listings of claims in the application with the following set of claims.

Claims 1-37 are currently pending. Claims 38-44 were previously withdrawn in response to a restriction requirement.

Claims 1-37 are hereby amended. Claims 45 to 50 are added as noted below.

1. (Currently Amended)     The method of synthesizing an oligonucleotide product for preferentially killing cancerous cells over non- cancerous cells comprising the steps of:

- (a) Using a first oligonucleotide comprising a nucleotide sequence, N, and with at least two CpG moieties; and
- (b) Covalently linking one or more units of an antimetabolite ((a)) prodrug for an antimetabolite covalently linked to the to said first oligonucleotide.

2. (Currently Amended)     The oligonucleotide method of claim 1, wherein the said antimetabolite is selected from the group consisting of 2'-deoxy-3'-thiacytidine, 3'-azido-3'-deoxythymidine, 2',3'-dideoxycytidine, 2',3'-didehydro-3'-deoxythymidine, 2',3'-dideoxyinosine, 5-fluoro-2' -deoxy uridine, 2-fluoro-9-b-D-arabinofuranosyladenine, I-B-Darabinofuranosylcytosine, 5-azacytidine, 5-aza-2'-deoxycytidine, 6-mercaptopurineriboside, 2-chlorodeoxyadenosine, and pentostatin.

3. (Currently Amended)     The method oligonucleotide of claim 1, wherein the said prodrug is a prodrug for the anitmetabolite is 2'- deoxy, 2', 2'- difluorocytidine.

4. (Currently Amended)     The method oligonucleotide of claim 1, wherein two of the said at least two CpG moieties are separated by a number of nucleotides, said number being selected from the group numbers 2, 5, and 9.

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5. (Currently Amended) The method oligonucleotide of claim 1, wherein one of said one or more units of an antimetabolite prodrug is linked 5' to the one of said at least two CpG moieties.

6. (Currently Amended) The method oligonucleotide of claim 1, wherein one of said one or more units of an antimetabolite prodrug is linked 3' to the one of said at least two CpG moieties.

7. (Currently Amended) The method oligonucleotide of claim 1, wherein one of said one or more units of an antimetabolite prodrug is linked 3' to the one of said at least two CpG moieties and one of said one or more units of an antimetabolite prodrug is linked 5' to the one of said at least ~~a second~~ two CpG moieties.

8. (Currently Amended) The method oligonucleotide of claim 1, wherein said one or more units of an antimetabolite prodrug is are linked to the said first oligonucleotide by a 3'-3' linkage.

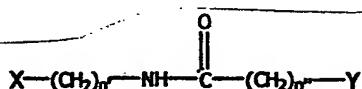
9. (Currently Amended) The method oligonucleotide of claim 1, wherein said one or more units of an antimetabolite prodrug is are linked to the said first oligonucleotide by a 5'-5' linkage.

10. (Currently Amended) The method oligonucleotide of claim 1, wherein said one or more units of an antimetabolite prodrug is are linked to the said first oligonucleotide by a 3'-5' linkage.

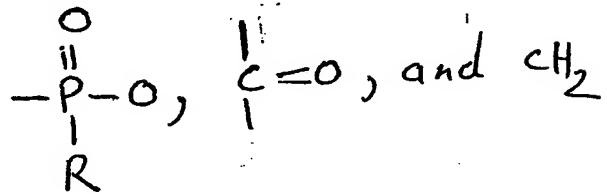
11. (Currently Amended) The method oligonucleotide of claim 1, wherein said one or more units of an antimetabolite prodrng is are eovalently linked to the said first oligonucleotide by a 5'-3' linkage.

12. (Currently Amended) The method oligonucleotide of claim 1, wherein one of said one or more units of an antimetabolite prodrng is eovalently linked at a position that is selected from 10 upstream s units from one of the said at least two CpO moieties, s being a whole number between 2 and 50.

13. (Currently Amended) The method oligonucleotide of claim 1, wherein the one of said one or more units of an antimetabolite prodrug is eovalently linked to the said first oligonucleotide by a linker having the formula.



wherein x and y are independently selected from



and R is selected from OH, S, a C<sub>1</sub>-C<sub>6</sub> alkyl, a C<sub>1</sub>-C<sub>6</sub> alkoxy, and NH.

14. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises contains at least one nucleotide having a ribose sugar moiety.

15. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises contains at least one nucleotide having a 2'-deoxyribose sugar moiety.

16. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises at least one 2'-halogen nucleotide.

17. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises at least one 2'-N-alkyl 2'-n-alkyl nucleotide wherein the alkyl has approximately having between about 1 and about 6 carbon atoms.

18. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises at least one nucleotide from the group 2'-O-alkyl 2'-o-alkyl nucleotide, one 2'-N-Alky1 2'-n-alky1 nucleotide, or one and 2'-O-halogen 2'-o-halogen and nucleotide, wherein the alkyl has approximately having between about 1 and about 6 carbon atoms.

19. (Currently Amended) The method oligonucleotide of claim 18, wherein the alkyl is o-methyl.

20. (Currently Amended) The method oligonucleotide of claim 1, wherein the oligonucleotide N comprises a plurality of nucleotides connected by covalent internucleoside linkages, wherein each of the linkages are is selected from the group

consisting of a phosphodiester linkage, a Cl-C6 alkoxy phosphotriester linkage, a phosphorothioate linkage and a phosphoramidate linkage.

21. (Currently Amended) A pharmaceutical composition comprising a therapeutically effective amount of the oligonucleotide made by a method of any of claims 1-20.

22. (Currently Amended) The oligonucleotide product of claim 21 wherein said pharmaceutically acceptable carrier is pharmaceutical composition comprises lipofectin as a carrier.

23. (Currently Amended) An The method of synthesizing an oligonucleotide product for preferentially killing cancerous cells over noncancerous cells comprising a motif represented by the formula: 5'PGXCG3' wherein P is a prodrug for an antimetabolite and X represents between 0 and 50 nucleotides. the steps of:

- (a) Using a first oligonucleotide with at least one CpG moiety and comprising a nucleotide sequence, X, having between ((0)) 2 and 50 nucleotides; and
- (b) Covalently linking a prodrug, P, for the antimetabolite 2'-deoxy, 2'-,2'-difluorocytidine to said first oligonucleotide so as to form PG, a second moiety of said oligonucleotide product.

24. (Canceled)

25. (Currently Amended) An The oligonucleotide of claim 23 method of synthesizing an oligonucleotide product for preferentially killing cancerous cells over noncancerous cells comprising the steps of:

- (a) Using a first oligonucleotide with at least one CpG moiety and comprising a nucleotide sequence, X, having between ((0)) 2 and 50 nucleotides; and
- (b) Covalently linking a prodrug, P, to said first oligonucleotide, wherein the metabolite P is selected from the group consisting of 2'-deoxy-3'-thiacytidine, 3'-azido-3'-deoxythymidine, 2',3'dideoxycytidine, 2',3'-didehydro-3'-deoxythymidine, 2',3'-dideoxyinosine, 5-fluoro-2'-deoxy uridine, 2-fluoro-9-b-D-arabinofuranosyladenine, 1-B-D-arabinofuranosylcytosine, 5-azacytidine, 5-aza-2'-deoxycytidine, 6-mercaptopurineriboside, 2-chlorodeoxyadenosine, and pentostatin.

26. (Currently Amended) The method oligonucleotide of claim of 23, where X is selected from the group consisting of 2, 5, and 9.

27. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises multiple nucleotides and the prodrug is covalently linked to one of the nucleotides by said covalent link of P is a 3'-3' linkage.

28. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises multiple nucleotides and the prodrug is covalently linked to one of the nucleotides by said covalent link of P is a 5'-5' linkage.

29. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises multiple nucleotides and the prodrug is covalently linked to one of the nucleotides by said covalent link of P is a 3'-5' linkage.

30. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises multiple nucleotides and the prodrug is covalently linked to one of the nucleotides by said covalent link of P is a 5'-3' linkage.

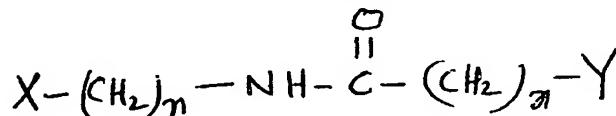
31. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises X contains at least one nucleotide having a ribose sugar moiety.

32. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises X contains at least one nucleotide having a 2'-deoxyribose sugar moiety.

33. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide X comprises at least one nucleotide from the group 2'-O-alkyl 2'-o-alkyl nucleotide, one 2'-N-Alkyl 2'-n-alkyl nucleotide, or one and 2'-O-halogen 2'-o-halogen and nucleotide, wherein the alkyl has approximately having between about 1 and about 6 carbon atoms.

34. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide X comprises a plurality of nucleotides connected by covalent internucleoside linkages, wherein each of the linkages are is selected from the group consisting of a phosphodiester linkage, a Cl-C6 alkoxy phosphotriester linkage, a phosphorothioate linkage and a phosphoramidate linkage.

35. (Currently Amended) The method oligonucleotide of claim 23, wherein the oligonucleotide comprises multiple nucleotides and the prodrug P is attached to at least one of the multiple nucleotides in X by a linker having the formula.



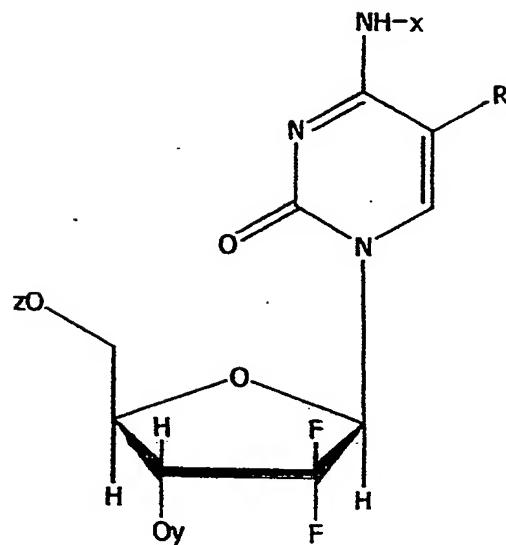
wherein x and y are independently selected from  $\overset{\text{O}}{\underset{\text{||}}{P}} - O$ ,  $\overset{\text{I}}{\underset{\text{|}}{C}} = O$ , and  $CH_2$

and R is selected from OH, S, a C<sub>1</sub>-C<sub>6</sub> alkyl, a C<sub>1</sub>-C<sub>6</sub> alkoxy, and NH.

36. (Currently Amended) A pharmaceutical composition comprising a therapeutically effective amount of the oligonucleotide made by the method of any of claims 23-35.

37. (Currently Amended) The oligonucleotide product of claim 36 wherein said pharmaceutically acceptable carrier is pharmaceutical composition comprises lipofectin as a carrier.

38. (Withdrawn) A compound having purity in excess of 98% by HPLC, having the formula:



wherein R is selected from the group consisting of H, a C1-C6 alkyl, a halogen, a C2-C6 alkenyl, and a C2-C6 alkynyl;

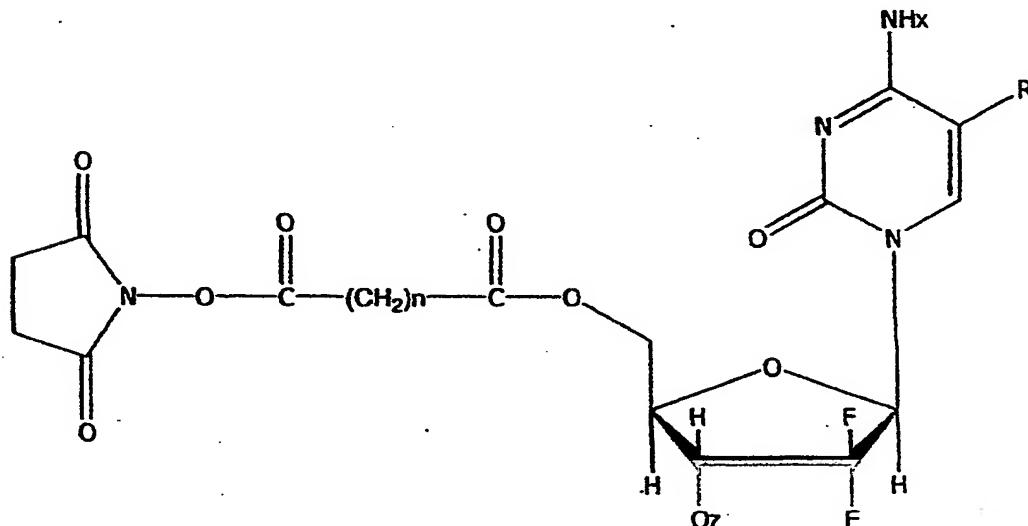
x is an amine-protecting group that is stable in oligonucleotide synthesis conditions; and

y, and z are each selected from H, a hydroxyl-protecting group that is stable in oligonucleotide synthesis conditions and a group that can be attached to a solid support.

39. (Withdrawn) The compound of claim 23, wherein the group that is attachable to a solid support has the formula O-C(=O)-M-C(=O)-NH-Spacer, where M is selected from the group consisting of succinyl, oxalyl, and hydroquinolynyl, and wherein the Spacer is selected from the

40. (Withdrawn) group consisting of a C1-C6 alkyl, ethyloxyglycol, and a combination of alkyl and ethyleneglycoxy.

41. (Withdrawn) A compound having the formula:



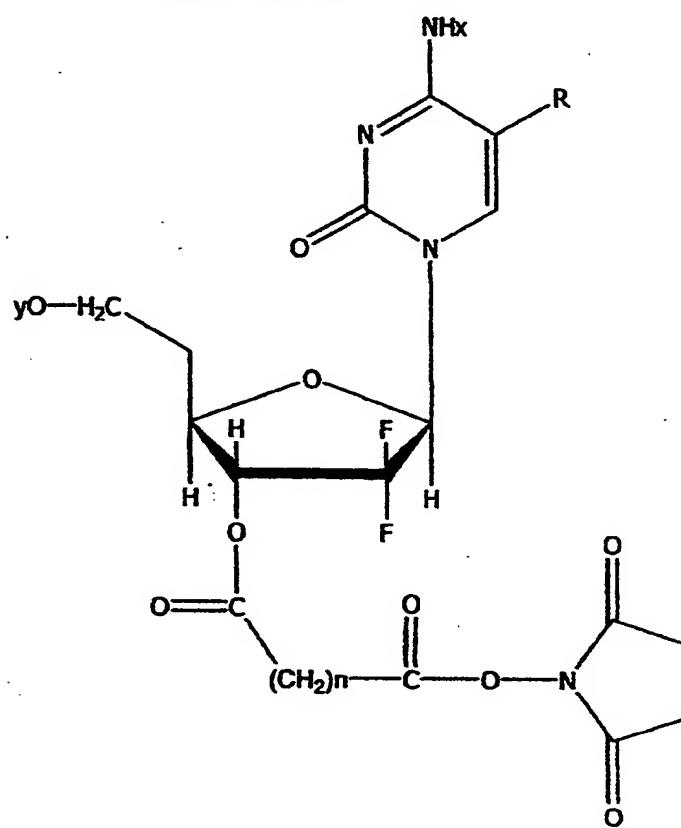
wherein R is selected from the group consisting of H, a C1-C6 alkyl, a halogen, a C2-C6 alkenyl, and a C2-C6 alkynyl;

x is an amine-protecting group that is stable in oligonucleotide synthesis conditions;

**z** is a hydroxyl-protecting group that is stable in oligonucleotide synthesis conditions; and

**n** is 2-20.

42. (Withdrawn) A compound having the formula:



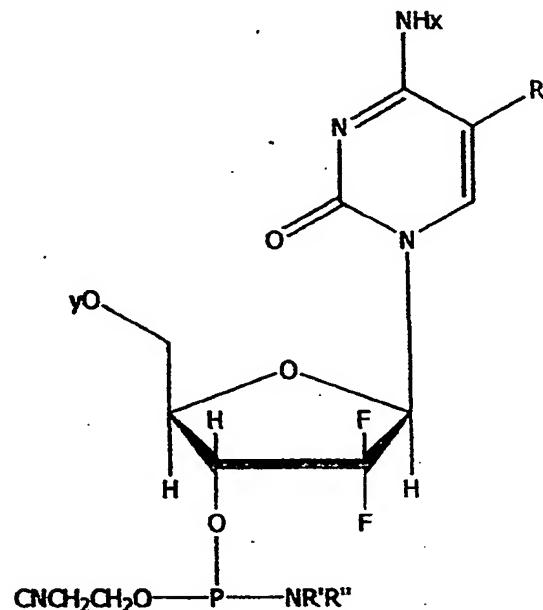
wherein **R** is selected from the group consisting of H, a C1-C6 alkyl, a halogen, a C2-C6 alkenyl, and a C2-C6 alkynyl;

**x** is an amine-protecting group that is stable in oligonucleotide synthesis conditions;

**z** is a hydroxyl-protecting group that is stable in oligonucleotide synthesis conditions; and

**n** is 2-20.

43. (Withdrawn) A compound having a purity in excess of 97% by HPLC, as shown by the formula:



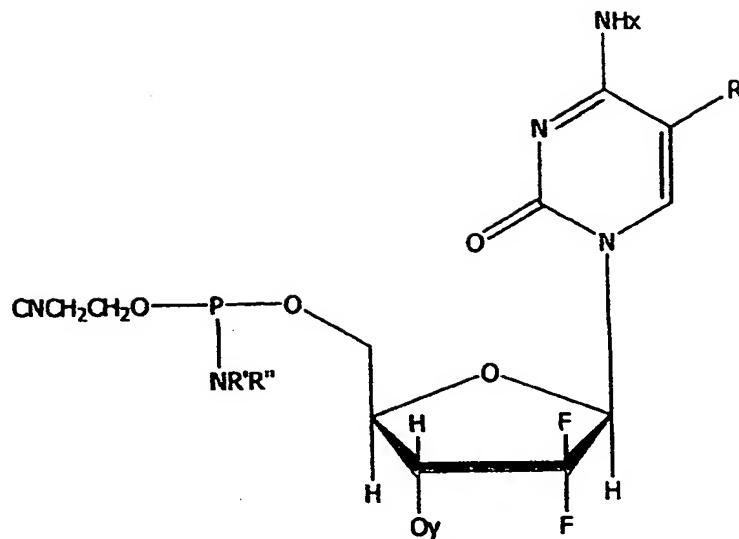
wherein  $\text{y}$  is a hydroxyl-protecting group that is stable in oligonucleotide synthesis conditions;

$\text{x}$  is an amine-protecting group that is stable in oligonucleotide synthesis conditions;

$\text{R}$  is selected from the group consisting of  $\text{H}$ , a C1-C6 alkyl, a halogen, a C2-C6 alkenyl, and a C2-C6 alkynyl; and

$\text{R}'$  and  $\text{R}''$  are independently selected from the group consisting of a C1-C6 alkyl and a C2-C6 cycloalkyl.

44. (Withdrawn) A compound having purity in excess of 97% by HPLC, and having the formula:



wherein y is a hydroxyl-protecting group that is stable in oligonucleotide synthesis conditions;

x is an amine-protecting group that is stable in oligonucleotide synthesis conditions;

R is selected from the group consisting of H, a C1-C6 alkyl, a halogen, a C2-C6 alkenyl, and a C2-C6 alkynyl; and

R' and R'' are independently selected from the group consisting of a C1-C6 alkyl and a C2-C6 cycloalkyl.

**(Claims 45-50 are newly added)**

45. (New) The method of claim 1, for preferentially killing cancerous cells in the colon, wherein said prodrug is antimetabolite dFC = 2'- deoxy, 2', 2'- difluorocytidine, dFCG represents the first CpG moiety of said first oligonucleotide, and N is GGACG.

46. (New) The method of claim 1, for preferentially killing cancerous cells in the colon, wherein said prodrug is antimetabolite dFC = 2'- deoxy, 2', 2'- difluorocytidine, dFCG represents the first CpG moiety of said first oligonucleotide, and N is GTGGAA.

47. (New) The method of claim 1, for preferentially killing cancerous cells in the colon, wherein said prodrug is antimetabolite dFC = 2'- deoxy, 2', 2'- difluorocytidine, dFCG represents the first CpG moiety of said first oligonucleotide, and N is GGACGTGGAA.

48. (New) The method of claim 1, for preferentially killing cancerous cells in the colon, wherein said prodrug is antimetabolite dFC = 2'- deoxy, 2', 2'- difluorocytidine, dFCG represents the first CpG moiety of said first oligonucleotide, and N is G GAGCTGGAACG.

49. (New) A pharmaceutical composition of claim 21 comprising a therapeutically effective amount of said oligonucleotide product.

50. (New) A pharmaceutical composition of claim 36 comprising a therapeutically effective amount of said oligonucleotide product.